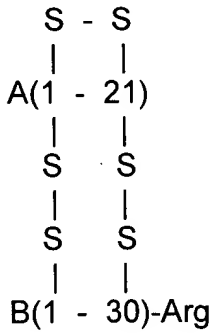
Petition for a One-Month Extension of Time and the required fee. Kindly enter the following amendment:

IN THE CLAIMS:

Please amend claims 21-22, 25-26, and 31 as follows:

21. (Twice Amended) A method for the preparation of a mono-Arg-insulin compound of formula II



(II)

in which A(1-21) and B(1-30) denote the A and B chains of human insulin and the -S-S-bridges are positioned as in insulin, which comprises:

(a) expressing in a bacterium a DNA molecule encoding a fusion protein which comprises a mini-proinsulin compound of the formula:

B(1-30)-Arg-A(1-31); - Type 7

(b) liberating said mini-proinsulin compound from said fusion protein;

(c) folding and forming disulfide bridges in said mini-proinsulin compound;

(d) incubating said mini-proinsulin compound with trypsin [at a pH of about 6.8 to produce mono-Arg-insulin, under conditions where no crystals are formed; followed by]; and

(e) precipitating the mono-Arg-insulin.

LAW OFFICES
FINNEGAN, HENDERSON,
FARABOW, GARRETT,
& DUNNER, L.L.P.
1300 I STREET, N. W.
WASHINGTON, DC 20005
202-408-4000

22. (Twice Amended) A method for the preparation of insulin which comprises:

(a) expressing in a bacterium a DNA molecule encoding a fusion protein which comprises a mini-proinsulin compound of the formula:

B(1-30)-Arg-A(1-21),

in which B(1-30) and A(1-21) denote the B and A chains of insulin;

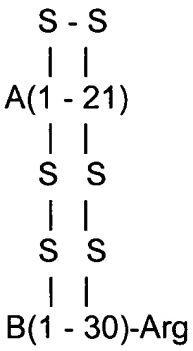
(b) liberating said mini-proinsulin compound from said fusion protein;

(c) folding and forming disulfide bridges in said mini-proinsulin compound;

(d) simultaneously incubating said mini-proinsulin compound with trypsin and carboxypeptidase B [at a pH of about 6.8 to produce insulin, under conditions where no crystals are formed; followed by]; and

(e) precipitating the insulin.

25. (Amended) A method for the preparation of a mono-Arg-insulin compound of formula II



(II)

in which A(1-21) and B(1-30) denote the A and B chains of human insulin and the -S-S-bridges are positioned as in insulin, which comprises:

(a) expressing in a bacterium a DNA molecule encoding a fusion protein which comprises

LAW OFFICES
FINNEGAN, HENDERSON, FARABOW, GARRETT, & DUNNER, L.L.P.
1300 I STREET, N. W.
WASHINGTON, DC 20005
202-408-4000

B(1-30)-Arg-A(1-21)

bonded via a bridging member,

- Met - Ile - Glu - Gly -Arg -,

to a peptide which stabilizes the fusion protein;

(b) liberating a mini-proinsulin compound from said fusion protein by cleaving the expressed fusion protein resulting from step (a) with cyanogen bromide;

(c) folding and forming disulfide bridges in said mini-proinsulin compound;

(d) incubating said mini-proinsulin compound with trypsin [at a pH of about 6.8 to produce mono-Arg-insulin, under conditions where no crystals are formed; followed by] ; and

(e) precipitating the mono-Arg-insulin.

26. (Amended) A method for the preparation of insulin which comprises:

(a) expressing in a bacterium a DNA molecule encoding a fusion protein which comprises

B(1-30)-Arg-A(1-21)

bonded via a bridging member,

- Met - Ile - Glu - Gly -Arg -,

to a peptide which stabilizes the fusion protein;

(b) liberating a mini-proinsulin compound from said fusion protein by cleaving the expressed fusion protein resulting from step (a) with cyanogen bromide;

LAW OFFICES
FINNEGAN, HENDERSON,
FARABOW, GARRETT,
& DUNNER, L.L.P.
1300 I STREET, N. W.
WASHINGTON, DC 20005
202-408-4000

(c) folding and forming disulfide bridges in said mini-proinsulin compound;

(d) simultaneously incubating said mini-proinsulin compound with trypsin and carboxypeptidase B [at a pH of about 6.8 to produce insulin, under conditions where no crystals are formed; followed by] ; and

(e) precipitating the insulin.

31. (Amended) A method for the preparation of insulin, without formation of substantial amounts of insulin Des-B30, comprising:

(a) expressing in a bacterium a DNA molecule encoding a fusion protein which comprises

B(1-30) - Arg - A(1-21)

bonded via a bridging member,

-Met-Ile-Glu-Gly-Arg-,

to a peptide which stabilizes the fusion protein;

(b) liberating a mini-proinsulin compound from said fusion protein by cleaving the expressed fusion resulting from step (a) with cyanogen bromide to produce mini-proinsulin;

(c) incubating the product formed in step (b) with sodium tetrathionate to form hexa-5-sulfonate;

(d) simultaneously incubating the S-sulfonate mini-proinsulin formed in step (c) with trypsin and carboxypeptidase [at a pH of about 6.8 under conditions where no crystals are formed]; and

(e) precipitating the insulin.

LAW OFFICES
FINNEGAN, HENDERSON,
FARABOW, GARRETT,
& DUNNER, L.L.P.
1300 I STREET, N.W.
WASHINGTON, DC 20005
202-408-4000

Please add the following new claims:

--33. A compound of the formula I

B(1-30)-Arg-A(1-21) (I)

wherein A(1-21) and B(1-30) denote the A and B chains of human insulin.

34. A nucleic acid sequence encoding the compound of formula I as claimed in claim 33.

35. A vector comprising the nucleic acid sequence of claim 34.

36. A host cell containing the nucleic acid sequence of claim 34.

37. A fusion protein comprising a compound of the formula I

B(1-30)-Arg-A(1-21) (I)

wherein A(1-21) and B(1-30) denote the A and B chains of human insulin, and wherein the compound is bonded via a bridging member

- Met - Ile - Glu - Gly - Arg -

to a peptide which stabilizes the fusion protein.

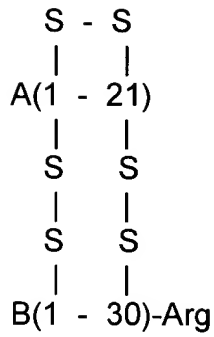
38. A process for the preparation a compound as claimed in claim 33, which comprises:

a) expressing a DNA sequence encoding the compound of the formula I in a bacterium; and

b) when the DNA sequence encodes a fusion protein, liberating the compound of formula I from the fusion protein.

39. A method for the preparation of a compound of the formula II

LAW OFFICES
FINNEGAN, HENDERSON,
FARABOW, GARRETT,
& DUNNER, L.L.P.
1300 I STREET, N.W.
WASHINGTON, DC 20005
202-408-4000



(II)

wherein A(1-21) and B(1-30) denote the A and B chains of human insulin and the -S-S- bridges are positioned as in insulin, comprising:

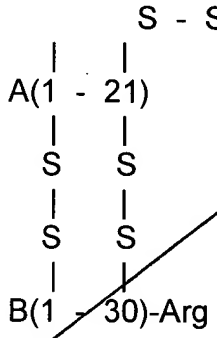
a) expressing a DNA sequence encoding the compound of formula I

B(1-30)-Arg-A(1-21) (I)

in a bacterium; and

b) cleaving the expressed compound of step (a) with trypsin.

40. A method for the preparation of a compound of the formula II



(II)

wherein A(1-21) and B(1-30) denote the A and B chains of human insulin and the -S-S- bridges are positioned as in insulin, comprising:

a) expressing a DNA sequence encoding the compound of formula I

B(1-30)-Arg-A(1-21) (I)

in a bacterium;

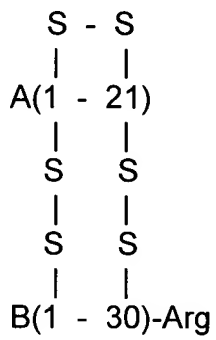
LAW OFFICES
FINNEGAN, HENDERSON,
FARABOW, GARRETT,
& DUNNER, L.L.P.
1300 I STREET, N. W.
WASHINGTON, DC 20005
202-408-4000

b) cleaving the expressed compound of step (a) with trypsin resulting in the compound of the formula II; and

(c) cleaving the resulting compound of step (b) with carboxypeptidase B.

41. The method of claim 40, wherein steps (b) and (c) are carried out in one vessel without having to isolate the intermediate compound of the formula II.

42. A method for the preparation of a mono-Arg-insulin compound of the formula II



(II)

in which A(1-21) and B(1-30) denote the A and B chains of human insulin and the -S-S-bridges are positioned as in insulin, which comprises:

(a) expressing a DNA sequence encoding a mini-proinsulin compound of the formula:

B(1-30)-Arg-A(1-31)

in a yeast; and

(b) cleaving said mini-proinsulin compound with trypsin.--

REMARKS

Claims 21-23, 25-27, 31, and 33-42 are currently pending in this application. Support for claims 33-44 can be found in the specification as a whole and pages 2-5

LAW OFFICES
FINNEGAN, HENDERSON,
FARABOW, GARRETT,
& DUNNER, L.L.P.
1300 I STREET, N. W.
WASHINGTON, DC 20005
202-408-4000